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Biochemistry of hyperthyroidism and hypothyroidism*

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Summary

The thyroid hormones act directly on mitochondria, and thereby control the transformation of the energy derived from oxidations into a form utilizable by the cell. Through their direct actions on mitochondria, the hormones also control indirectly the rate of protein synthesis and thereby the amount of oxidative apparatus in the cell. A rationale for the effects of thyroid hormone excess or deficiency is based upon studies of the mechanism of thyroid hormone action. In hypothyroidism, slow fuel consumption leads to a low output of utilizable energy. In hyperthyroidism, rapid fuel consumption leads to a high energy output, but as efficiency decreases, the utilizable energy produced decreases. Many of the chemical and physical features of these diseases can be reduced to changes in available energy.

Introduction

Excess or deficiency in the amount of thyroid hormones in humans produces clinical and chemical manifestations that involve a number of organ and metabolic systems. Variations of thyroid hormone concentrations *in vivo* change oxygen consumption, temperature regulation, growth and development, the response to other hormones, nerve function, and the metabolism of proteins, fats, carbohydrates, nucleic acids, vitamins, and inorganic anions and cations. On the other hand, thyroxine and triiodothyronine are relatively simple molecules, and their small size and limited number of reactive groups suggest either that the variety of the effects they produce are due to a few types of primary interactions at the molecular level, or that the hormones are changed in the body to analogues each having a different and specific physiologic effect. The preponderance of evidence at present supports the first hypothesis. It seems feasible therefore to attempt to reduce the complex pathologic pictures to subcellular phenomena and their consequences.

Recent advances in the understanding of where and how the thyroid hormones act in the cell support a simplification of thyrotoxicosis and hypothyroidism, although our understanding is not as yet so far advanced as to permit a final 'explanation' of the diseases in molecular terms. A brief history of the evolution of studies on the mechanism of thyroid hormone action serves to outline the present state of knowledge, the areas in which future advances may be made, and a basis for a rationale of hyperthyroidism and hypothyroidism.

Actions and effects of thyroid hormones

Ever since Magnus-Levy (1895) showed that the thyroid gland controlled the rate of oxygen consumption in mammals, attention has been fixed on oxidative processes as a target of the hormone. Kendall (1929) showed the structure of thyroxine and suggested the hormone might be a component or coenzyme of an oxidative enzyme, undergoing a redox cycle between the phenol and semiquinone forms. No evidence has as yet been found to support Kendall's hypothesis conclusively. In the years 1940–50 it became clear that 90% or more of the cell's oxygen was consumed via processes occurring in mitochondria, and experiments were done with thyroid hormones *in vivo* and *in vitro* to determine their effects on mitochondria.

One should differentiate, in considering these studies, between *actions* of the hormones and *effects* of the hormones. *Actions* may be defined as those functional or structural changes that are primary and depend upon the presence of the hormone at a site where it interacts with a molecule in the cellular apparatus. Because hormones are effective in small amounts we may assume that their primary molecular interactions are reversible, so that the hormones are not

**Abbreviations:* L-T_4, L-thyroxine; L-T_3, L-triiodothyronine; Triac, triiodothyroacetic acid; ATP, ADP and AMP, adenosine tri-, di- and mono-phosphate; P_i, inorganic phosphate; NADH and NADPH, reduced nicotinamide-adenine dinucleotide and dinucleotide phosphate; DNP, 2,4-dinitrophenol; BMR, basal metabolic rate (O_2 consumption).

used up. *Effects* may be defined as those functional, structural, or compositional changes that are secondary and do not depend upon the presence of the hormone; they should not be reversed if the hormone is removed after acting. The differentiation between actions and effects makes no judgement on their relative importance in the cell. The thyroid hormones are peculiarly suitable for the resolution of primary actions from secondary effects, because their iodine moieties can be used experimentally as a tracer for quantitative analysis. Methods for detecting other hormones not possessing this useful property are less specific or more tedious.

As will be detailed below, the thyroid hormones were shown to affect mitochondria as 2,4-dinitrophenol did: both agents increased mitochondrial respiration, and the energy liberated was transformed into heat rather than into the normal utilizable form, the high-energy phosphate bond. This toxic, catabolic, energy-wasting effect served as a rationale for thyrotoxicosis (Hoch, 1962a), but not for the anabolic energy-conserving effects that the smaller doses of thyroid hormones exerted in euthyroid or hypothyroid subjects (Hoch, 1962b). Nor was hypothyroidism made more understandable by the 'uncoupling' hypothesis. Accordingly, attention was directed away from the mitochrondrion in the search for the mechanism.

In the early 1960s the groups of Tata and of Sokoloff showed that thyroid hormones affected protein synthesis. It had been demonstrated earlier by Dutoit (1952) that protein was synthesized abnormally slowly in the livers of hypothyroid rats. L-T_3 given *in vivo* accelerated the synthesis of proteins by ribosomes after about 48 hr after injection; the doses necessary were small and physiologic, smaller than those producing uncoupling in mitochondria, and the effect of the hormone was obviously anabolic (see Tata, 1967). Puromycin and actinomycin D, agents that block protein synthesis by acting on nucleic acids, blocked the calorigenic action of thyroid hormones (Tata, 1963; Weiss & Sokoloff, 1963). No changes were observed in mitochondrial respiratory control after hormone injection (Tata *et al.*, 1963). Respiratory acceleration could be demonstrated in mitochondria 70–90 hr after hormone injections, but it was due to increases in the number of depleted respiratory assemblies in the mitochondria of hypothyroid rats (Tata *et al.*, 1963; Roodyn, Freeman & Tata, 1965), and so represented the specific results of earlier protein synthesis. Increases in nuclear RNA-metabolism were shown early (3–16 hr) after hormone injection (Tata & Widnell, 1966), then increases in ribosomal RNA-content and aggregation (about 40 hr) (Tata, 1967). However, although all these phenomena showed an important relationship between the thyroid hormones and the processes supplying information to and controlling the rate of protein synthesis, they did not show the primary locus of hormone action. When L-T_3 was added to isolated nuclei, RNA-metabolism was not stimulated (Widnell & Tata, 1963; Tata & Widnell, 1966; Sokoloff, Francis & Campbell, 1964). Tata's conclusions are diagrammed in Fig. 1.

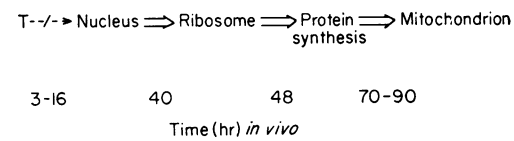

FIG. 1. Sequence of events after injecting hypothyroid rats with thyroid hormone (T = L-T_3), according to Tata and co-workers.

The studies of Sokoloff (Sokoloff & Kaufman, 1959, 1961; Sokoloff *et al.*, 1963, 1964) have recently drawn attention back to the mitochondrion as a site of action of the hormone (Fig. 2).

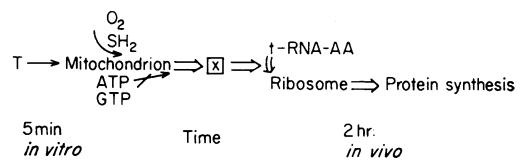

FIG. 2. Sequence of events after injecting hypothyroid rats with thyroid hormone, or after addition of thyroid hormone to mitochondria (T = L-T_3), according to Sokoloff and co-workers.

Adding L-T_3 to a homogenate *in vitro* stimulated ribosomal synthesis of proteins. The processes whereby t-RNA-amino-acyl complexes interacted with the ribosomes were the locus of the stimulation. Mitochondria oxidizing a substrate were necessary, and they apparently produced a substance that accelerated the ribosomal translation; adding ATP, GTP or glutathione did not replace the effect of hormone-treated mitochondria. What it is that mitochondria produce to control ribosomal protein synthesis is not yet clear; studies by Bronk (1963) have suggested that mitochondrial non-phosphorylated, high-energy intermediates may support protein synthesis.

Our recent studies have shown that L-T_4 injected *in vivo* can act rapidly and directly on

mitochondria (Hoch, 1968a). Bronk (1966) has also shown a very short latent period for L-T_3. In hypothyroid rats, a subcalorigenic dose of L-T_4 (at least fifty times less than those used to stimulate protein synthesis) partly corrected the excessive respiratory control in liver mitochondria 3 hr after injection (Hoch, 1966). A larger dose did the same when the rats were killed 2 min after injection (Hoch, 1967, 1968b). The hormone content of mitochondria, as measured by the total iodine or the butanol-extractable iodine, was 20% of normal in untreated hypothyroid rats, and rose progressively with the functional changes up to 3 hr after injection (Hoch, 1967; Dillon & Hoch, 1968); the amount of hormone was one to five molecules per respiratory assembly in the treated rats, and about 50 μM in the mitochondrion (a concentration effective *in vitro* in Sokoloff's experiments). This early or instantaneous action of the hormone was completely reversed when bovine serum albumin was added to the mitochondria, and the greater part of the hormone was thereby removed (Hoch & Motta, 1968). The features of the reversibility of the functional changes demonstrated the following: (a) that the hormone acted directly on mitochondria, but perhaps through an intermediate or synergistically with endogenous mitochondrial components; (b) that synthesis of a mitochondrial protein was not involved; and (c) that observations (Tata *et al.*, 1963) that the hormone produced new mitochondrial enzymes demonstrated an effect and not an action of the hormone, because those workers had routinely added bovine serum albumin to their assay mixtures and so had observed only the late irreversible effects of L-T_3.

At the present time, the actions and the effects of the thyroid hormones appear to be related as in Fig. 3. Low doses of the hormone act

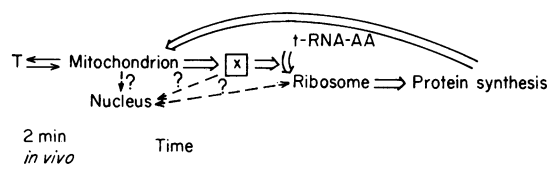

FIG. 3. Sequence of events after injecting hypothyroid rats with thyroid hormone (T = L-T_4), according to Hoch and co-workers.

rapidly or instantaneously, and reversibly, on mitochondria (only liver mitochondria have so far shown unmistakable results of treating hypothyroid rats with the hormone). The functional changes in mitochondria accelerate, by a process not yet clear, the rate of translation of t-RNA-amino-acyl complexes by ribosomes to synthesize proteins. Among the proteins synthesized are the enzymatic components of the mitochondrial respiratory chain. The nucleus is also involved early, but the relation between the rises in nuclear RNA-metabolism and the earlier changes in mitochondrial function, as well as the later changes in ribosomal metabolism and mitochondrial composition, are also not yet finally defined. Thus, mitochondria show changes in function dependent upon the hormone's presence or absence, and changes in enzyme content secondary to the alterations in protein synthesis that depend ultimately upon the functional changes.

It now appears profitable to consider mitochondrial respiration and energy-transformation as the loci at which the thyroid hormone normally acts, and at which excess or deficiency in hormone content exerts its primary action.

Early functional changes

Mitochondria may be regarded as energy transforming or transducing machines performing oxidative phosphorylation: liberating energy by oxidizing a substrate, and transforming this energy into a chemically utilizable form for endergonic reactions, the high-energy phosphate bonds of adenosine-triphosphate (ATP). The molecular events of this process are the subject of intensive investigation (see Lehninger, 1964; Racker, 1965) but are as yet incompletely understood. Oxidative phosphorylation is measured by the rate of oxygen consumption (energy input per unit of time) and the efficiency of energy transfer, the P:O ratio (utilizable energy output per energy input). The amount of work the machine can do per unit of time (the utilizable energy output per unit of time) is in physical units, *power*. The useful output of the mitochondrion, its oxidative power, consists of high-energy phosphate bonds.

Physiologically, the most important feature of oxidative phosphorylation is that it is *self-regulating*. The rate of output of high-energy phosphate bonds controls the rate of input, i.e. the consumption of oxygen and the oxidation of substrates. ADP controls the rate of oxidation; ADP accepts the high-energy phosphate groups from a hypothetical mitochondrial intermediate to form ATP. In the absence of added ADP, or in the absence of any agency removing the terminal phosphate groups of ATP to form ADP, mitochondria oxidize substrates very slowly (State 4) as in Fig. 4. Addition of ADP, or the presence of an enzyme system hydrolysing ATP to ADP, increases the rate of oxidation markedly

(State 3) as in Fig. 5. The *respiratory control* ratio is the ratio of the rates of respiration in State 3:State 4. The mitochondria in resting living cells function as if little ADP were available; that is, cells respire in State 4, in a controlled condition in which large demands for utilizable energy (production of ADP) can be met with bursts of high oxidative activity (see Hoch, 1968b).

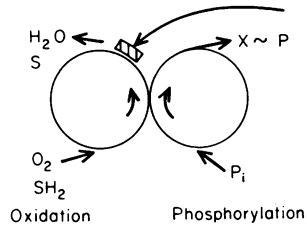

FIG. 4. Normal mitochondrial oxidative phosphorylation in State 4, with no ~ P-acceptor. The − X ~ P group exerts a braking effect on the oxidation cycle.

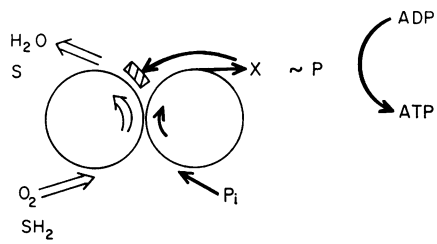

FIG. 5. Normal oxidative phosphorylation in State 3, in the presence of ADP. The free −X group exerts no braking effect on the oxidation cycle, and oxidation is accelerated.

The efficiency of phosphorylation, the P:O ratio, is the number of moles of high-energy phosphate produced per gram atom of oxygen consumed. This ratio is 3 for most compounds oxidized via diphosphopyridine-nucleotide-dependent dehydrogenases. The translation of the energy liberated by oxidation into phosphoryl-bond energy has been termed 'coupling' by mechanical analogy (Loomis & Lipmann, 1948). Phosphorylation can be decreased or abolished selectively without diminishing mitochondrial oxidations. Physical agencies (e.g. heat or hypotonicity) or a variety of chemical agents (the classical one is 2,4-dinitrophenol) 'uncouple' oxidative phosphorylation, and decrease the phosphorylation quotient by decreasing its numerator. Many of the chemical agents also uncouple *in vivo* when administered to normal animals, i.e. the mitochondria subsequently isolated from the treated animal phosphorylate with decreased efficiency.

The *efficiency* of oxidative phosphorylation also controls the oxidative rate. Uncoupling raises the respiratory rate markedly (State 3u), and depresses the respiratory control ratio (Fig. 6); addition of ADP does not then accelerate the already rapid respiration. Low concentrations of uncoupling agents, insufficient to decrease the efficiency of phosphorylation measurably, also increase oxygen consumption and lower respiratory control; this is termed 'loose coupling' (Fig. 7). The mechanism that ordinarily limits the rate of respiration when ADP is absent (i.e. when —X~P remains undischarged) now permits both rapid respiration and transfer of energy.

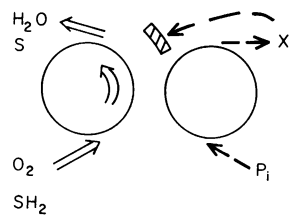

FIG. 6. Uncoupled oxidative phosphorylation in the absence of ADP (State 3u). The −X group exerts no braking effect on the oxidation cycle. Adding ADP will not accelerate oxidation. Phosphorylation is abolished (efficiency = 0).

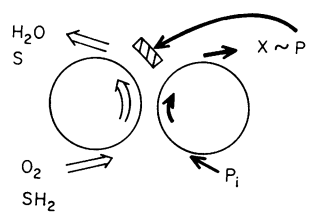

FIG. 7. Loose-coupled oxidative phosphorylation in the absence of ADP (State 4). The − X ~ P group now exerts no braking effect on the oxidation cycle. Adding ADP will not accelerate oxidation. Phosphorylation is at almost normal efficiency.

L-T_4 and L-T_3 can act like uncoupling agents in many respects. Large doses *in vivo* and high concentrations *in vitro* uncouple oxidative phosphorylation (see Hoch, 1962b; Hoch & Lipmann, 1954; Maley & Lardy, 1953; Martius & Hess, 1952). By either route, the hormone accelerates the State 4 oxidation of most substrates and depresses the respiratory control. Much smaller doses or concentrations also lower respiratory

control by raising State 4 oxidation, but they do not depress the P:O ratio (see Fig. 7), nor interfere with the inhibitory action of oligomycin (Hoch, 1968b), an agent specific for phosphorylating respiration.

Mitochondria from hypothyroid animals, that contain only 20% of the normal amount of thyroid hormone, are 'over-coupled', respiring too slowly in State 4 with excessive respiratory control (Maley & Lardy, 1955), as in Fig. 8. It

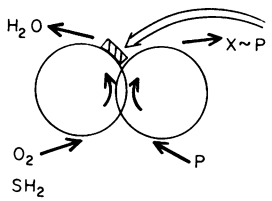

FIG. 8. 'Over-coupled' oxidative phosphorylation in State 4. The $-X \sim P$ group now exerts a greater than normal braking effect on the oxidation cycle. Adding ADP will accelerate oxidation to (almost) normal levels. Phosphorylation is normal.

seems that the optimal amount of L-T_4 is necessary to poise mitochondrial energy-transfer between inertia and inefficiency.

Consideration of *oxidative power* as an index of the performance of the mitochondrial mechanism offers a basis for rationalizing many of the diverse effects of the thyroid hormones. In Fig. 9, oxidative power is viewed as a resultant of changes in the rate of oxidation and/or the efficiency of energy transfer (Hoch, 1962a, 1968a). The rate of energy consumption (the

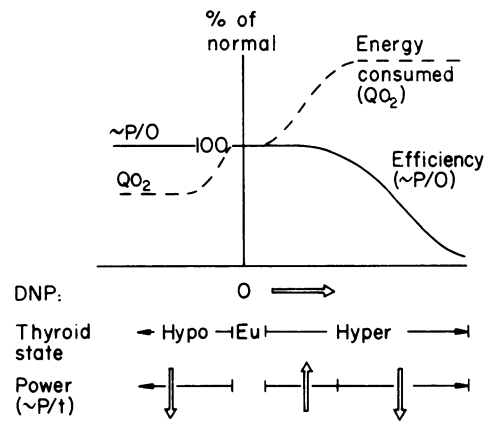

FIG. 9. Fuel consumption (QO_2), efficiency ($\sim$ P:O), and power ($\sim$ P:t) of the mitochondrion as a function of the concentration of DNP or thyroid hormone (from Hoch, 1968a).

QO_2) and the efficiency of the machine (the $\sim$P:O ratio) are plotted as a function of the concentration of an agent that can uncouple, e.g. DNP or thyroid hormone. It will be seen that low concentrations of DNP or thyroid hormone stimulate energy consumption before efficiency is depressed; the result is increased power. Higher concentrations uncouple and depress efficiency. Power therefore is decreased, and it is this catabolic effect that is seen in thyrotoxicosis. In hypothyroid subjects, mitochondrial energy consumption is depressed below normal, but efficiency is normal. Power is therefore decreased. Treatment with L-T_4 raises energy consumption, but does not affect efficiency: power is restored to normal, an anabolic action. DNP has been shown to possess such an anabolic action as well, in accelerating protein synthesis *in vitro* (Sokoloff & Kaufman, 1961).

However, the differences between the thyroid hormones and the uncoupling agents should be stressed at this point: certain effects of the thyroid hormones are biologically specific. Agents like DNP do not relieve all the defects seen in hypothyroid subjects, nor do they stimulate growth and development, nor do they induce metamorphosis in *Anura*. The specificity must reside in the differences between their actions at the mitochondrial level. The mechanisms of action of thyroxine and DNP are known to differ: thyroxine makes mitochondria swell and DNP does not; and thyroxine and DNP act synergistically on mitochondrial respiration, not additively.

At least two factors may govern the effect of thyroid hormones: the thyroid state of the organism (presumably a function of the concentration of thyroid hormone in contact with the target sites) and the additional amount of the administered hormone reaching the target. Thus, thyroid hormone given to a hypothyroid subject raises oxidative metabolic power to normal levels; similar dosage in a euthyroid subject increases power above normal; higher dosage decreases power.

Late compositional changes

The functional changes in mitochondrial respiration discussed in the previous section involve *specific* respiratory activity, in the sense of the rate of respiration per amount of respiring mass. The thyroid hormones also affect the *total* respiratory mass in the mitochondrion. They control, through protein synthesis, the amount of respiratory enzymes per gram of total protein in the mitochondrion. In hypothyroidism, the rate of protein synthesis is lower than normal.

Mitochondria from hypothyroid rats contain half as much cytochrome b, c, and $a+a_3$ as normal mitochondria (Maley, 1957; Kadenbach, 1966) and perhaps less flavoprotein catalysts (Rivlin & Langdon, 1966). But, paradoxically, there is twice the normal amount of pyridine nucleotide coenzymes. Conversely, in mild hyperthyroidism, with its raised rate of protein synthesis, the content of cytochromes and flavoproteins is high, and of pyridine nucleotides, low (Kadenbach, 1966; Maley & Lardy, 1955).

At first glance, these compositional differences seem to reflect or account for the abnormal BMRs in thyroid disease. However, the situation is not so clear. The flavoproteins and cytochromes have turnover numbers, and are present in such amounts in mitochondria, that they do not control or limit oxidation in the respiratory chain. The 'bottle-neck' in respiration (actually in electron-transport) appears to be at or near the pyridine nucleotide end of the chain (Chance, 1965), and there is evidence that the substrate-dehydrogenase interactions are involved (Klingenberg, 1963). The pyridine nucleotides, however, change in amount in a direction opposite to the changes in BMR, under thyroid hormone influence. Thus, while the hormone seems to control the maximal *capacity* for oxidation in mitochondria, it also controls the *specific* activity of respiration in the chain, and the latter accounts for the rate of oxygen consumption in the resting subject (Ernster & Luft, 1964). The importance of capacity for oxidation in processes involving large demands for energy, however, should not be minimized.

The resting hyperthyroid subject thus consumes oxygen faster because the mitochondria in his tissues consume oxygen faster in State 4. The term 'hypermetabolism' in this connection is a misnomer, for while oxidation is increased, the metabolic processes of phosphorylative energy transfer are normal or decreased. This kind of hyperoxidation arises from the presence of excess amounts of thyroid hormones in the mitochondria. Not all organs consume more oxygen in thyrotoxic subjects; the brain, spleen and testes do not. The mitochondria of brain, spleen and testes do not swell in the presence of thyroid hormones, in contrast to those from other tissues (Tapley & Cooper, 1956), but it is not known if their iodine contents are high or normal in thyrotoxic subjects. Another kind of hyperoxidation may arise from the late adaptive increases in mitochondrial enzyme content, but it is presumably fully efficient and should persist for some time even when the amount of thyroid hormone in the mitochondria becomes normal.

The increased rates of oxidation are accompanied by increased production of heat per unit of time even when efficiency is normal, and relatively more heat than utilizable energy will be evolved as the efficiency of energy transfer decreases. In the most extreme forms of thyrotoxicosis, where more complete uncoupling might occur, heat production leads to hyperpyrexia (as in thyroid crisis; see below). Lesser degrees of heat production are compensated by sweating, vascular changes, dilation, flushing, and resultant tachycardia, increased cardiac stroke volume and pulse pressure. Such changes may in part be mediated through the action of the hormones of the adrenal medulla (see pp. 357–8). Compensation may be precarious, and the excessive demands imposed by relatively slight increases of external temperature, the rise of heat production during muscular exercise, or administration of agents with uncoupling properties may have drastic consequences. Increased tissue heat emphasizes the increased metabolic demands and further accelerates oxidative rate. Weight loss and wasting of both fat and lean body mass (Wayne, 1960) occur without any losses in appetite.

Conversely, the hypothyroid subject consumes oxygen more slowly than normally because his mitochondria respire slowly in State 4. Both because of thyroid hormone deficiency and respiratory enzyme depletion, 'hypometabolism' is here not a misnomer, phosphorylative metabolism proceeding at a low rate because of the diminished liberation of oxidative energy. Decreased rates of oxidation produce less heat, and extremes result in hypothermia (see myxoedema coma, below). Demands for increased heat production, as in acclimatization to cold environments, are met poorly by hypothyroid subjects. Responses to the hormones of the adrenal medulla are subnormal. It is of some interest that clinical investigators (Selenkow & Marcus, 1960) have commented upon the 'apathetic' hyperthyroidism seen in older patients. It has features similar to those of hypothyroidism; on our basis, both are reflections of the decreased capacities for the production of utilizable energy.

Alterations in metabolism

Certain features of clinical and experimental hyperthyroidism and hypothyroidism may be considered as manifestations of changes in the transformation of energy. More information is available on the correlation of manifestations and basic changes in hyperthyroidism than in hypothyroidism. The following are those areas of energy utilization best studied at present.

Protein metabolism

Thyroid hormones control protein synthesis and breakdown. The effect of administered thyroid hormones depends upon the thyroid status of the subject. Low doses of thyroxine stimulate protein synthesis, high doses depress it. L-T_3 decreased protein synthesis in euthyroid humans, but the same dose raised the low rate of protein synthesis to normal levels in myxoedematous patients (Crispell, Parson & Hollifield, 1956). In hypothyroid rats, 5–10 μg of T_4 daily increased protein synthesis (Karp & Stetten, 1949; Rupp, Paschkis & Cantarow, 1949), but in either hypothyroid or normal animals, 50–100 μg decreased or abolished synthesis (Rupp *et al.*, 1949; Sokoloff & Kaufman, 1959). Hyperthyroid humans have subnormal amounts of parenchymal protein in liver biopsies (Nikkilä & Pitkänen, 1959). Consistent with the decreased synthesis of peptide linkages, the concentrations of free amino acids in blood, liver and muscle are elevated in thyrotoxic rats (Crispell *et al.*, 1956; Friedberg & Greenberg, 1947). Rat muscle (Ferrini, Perroni & Bestagno, 1959) and human cells growing in culture (Leslie & Sinclair, 1959) incorporate amino acids into proteins more slowly in the presence of added thyroxine. Sokoloff & Kaufman's (1961) studies have demonstrated *in vitro* that the apparently conflicting effects of T_4 upon protein synthesis are in reality biphasic hormonal effects.

Lipid metabolism

Thyroid hormones control the rates of lipid synthesis, oxidation and mobilization. Biphasic effects of thyroid hormone have been shown on lipid synthesis (Fletcher & Myant, 1960); 20 μg of thyroxine increased the synthesis of cholesterol from acetate by the cell-free fractions of rat livers, while 30–50 μg decreased the synthesis. Fatty acid synthesis was decreased at all these levels of dosage.

Thyroid-treated rats and humans synthesized cholesterol more rapidly than normal (Kritchevsky, 1960), and the hypocholesterolaemia of thyrotoxicosis, anomalous in the face of increased synthesis, was ascribed to the hormonal stimulation of cholesterol excretion (Rosenman, Byers & Friedman, 1952). Increased rates of cholesterol and fatty acid synthesis have been observed in hormone-treated rats and in their tissue slices (Karp & Stetten, 1949; Dayton *et al.*, 1960). On the other hand, decreased rates of cholesterol synthesis were observed in liver homogenates (Scaife & Migicovsky, 1957) and cell-free preparations (Fletcher & Myant, 1962) from thyrotoxic rats. The cholesterol and neutral fat contents of the livers of thyroxine-treated rats (Handler, 1948), and the fat content of human bodies (Wayne, 1960) were below normal in hyperthyroidism. Thyroidectomy also decreased the rate of cholesterol synthesis (Boyd, 1959); the low rate of synthesis of cholesterol and fatty acids in myxoedematous subjects was raised to normal by thyroid hormones (Lipsky *et al.*, 1955) However, the control of lipid synthesis by thyroid hormones has also been observed in tissue preparations that were supposed to be free of mitochondria (Fletcher & Myant, 1960).

The defects and increases in lipid synthesis might be ascribed to changes in the availability of ATP at various steps in the process, even if mitochondria were not present in the lipid-synthesizing preparations. Thus, the effects of thyroid hormones have been laid to alterations in the supply of acetyl-coenzyme A, or further along the synthetic path, in the conversion of acetate to cholesterol, fatty acids and CO_2 (Dayton *et al.*, 1960); both require ATP. Coenzyme A concentrations do vary in the tissues of thyrotoxic animals, being low in hyperthyroid rat livers (Fraenkel-Conrat & Greenberg, 1946) and in hyperthyroid humans (Gershberg & Kuhl, 1950), and rising above normal when thyroidectomized rats are treated with thyroxine, apparently via increased availability of ATP (Tabachnick & Bonnycastle, 1954).

The fatty acid oxidases are in mitochondria. Thyroid hormones can act directly on lipid oxidations by controlling ATP production, since fatty acid activation requires ATP prior to oxidation. Thyroxine treatment accelerated fatty acid oxidation in rat heart homogenates (Deitrich & Smith, 1960), earlier than it raised the basal metabolic rate (Abelin & Kürsteiner, 1928). Bacterial oxidation of cholesterol was increased by added thyroid hormones (Wainfan & Marx, 1955). Hypothyroidism decreased fatty acid oxidation and ATP production from fatty acids in the hearts of dogs.

Thyroid hormones also control fatty acid concentrations in tissues through the rate of the mobilization of fatty acids from adipose tissue, in conjunction with the action of other hormones. T_3 and Triac raised serum concentrations of unesterified fatty acids within 6 hr in humans (Rich, Bierman & Schwartz, 1959) and enhanced their release from adipose tissue and their removal from serum in dogs. These thyroid effects are facilitations of other stimuli, particularly epinephrine (Jeanrenaud, 1961), which ordinarily liberate fatty acids (Schwartz & Debons, 1959). Epinephrine-induced mobilization of fatty acids *in vivo* requires optimal

thyroid function, while thyroid hormone alone does not release fatty acids from adipose tissue *in vitro* (White & Engel, 1958). Hypothyroidism prevents epinephrine-induced mobilization. In hypopituitary monkeys epinephrine injection liberated no fatty acids; thyroid-stimulating hormone partly restored, and small doses of T_3 fully restored, the normal fat-mobilizing response to epinephrine (Goodman & Knobil, 1959). Hyperthyroidism exaggerates the epinephrine effect on fat pads *in vitro* (Debons & Schwartz, 1961) and hypothyroidism abolishes it. The fat-mobilization effects of insulin are similarly affected by the thyroid state, insulin releasing six times more free fatty acids from the adipose tissue of T_4-injected rats than normally (Hagen, 1960). These interdependences of hormone effects may reflect the facts that fat mobilization depends upon the production of cyclic-3′,5′-AMP, and that cyclic-3′,5′-AMP is formed only from ATP; the thyroid hormones control ATP production, and it may be speculated—in the absence of definite information to date—that the ATP supply, as well as the hormone-controlled activity of adenyl cyclase, controls cyclic-3′,5′-AMP production.

Carbohydrate metabolism

Thyroid hormones control the rates of glycogen synthesis and breakdown, and of hexose oxidation.

Thyroxine has a biphasic effect on glycogen synthesis. Low doses of thyroxine increased glycogen synthesis in rat diaphragms either *in vivo* or *in vitro*, and higher doses reversed this effect (Wertheimer & Bentor, 1953). *In vivo* injections of thyroid-stimulating hormones (in normal but not in thyroidectomized animals), or of 20–30 μg of L-thyroxine, increased glycogen synthesis, whereas 100–200 μg of L-thyroxine decreased synthesis below normal rates. *In vitro*, incubation of 2 μg of L-thyroxine with normal rat diaphragms increased glycogen synthesis while higher amounts either did not stimulate, or irregularly depressed, synthesis. Both the *in vitro* and the *in vivo* effects depended upon incubation of the diaphragms in homologous rat serum, which may have involved thyroxine-binding or lipid-binding. The lack of a measurable rise in oxygen consumption, however, indicates caution in accepting this system as one depending simply on ATP-supply.

Most of the available evidence indicates decreased synthesis of glycogen in thyrotoxicosis. Glycogen synthesis was decreased in hyperthyroid humans and rabbits (Coggeshall & Greene, 1933; Mirsky & Broh-Kahn, 1936). The glycogen contents of liver and muscle were markedly diminished in hyperthyroid subjects, especially the metabolically active forms of glycogen (Chilson & Sacks, 1959), but this, of course, may also depend on increased breakdown. Consistent with a decrease in synthesis is the fact that both liver and muscle (where the major portion of glycogen synthesis proceeds) showed decreased contents of ATP after thyroid hormones were administered (Chatagner & Gautheron, 1960; Berg, 1937). Increases in synthetic rates have also been observed, however, after single doses (Sternheimer, 1939) or more prolonged hormone treatment followed by liver perfusion with large amounts of glucose (Burton, Robbins & Byers, 1958).

The hyperglycaemic effect of epinephrine, mediated through the increased formation of cyclic-3′,5′-AMP and the subsequent activation of phosphorylase, depends upon the thyroid state, and administered thyroid hormone produces biphasic effects. A small dose of T_4 raised the hyperglycaemic effect of injected epinephrine whereas in rabbits fed 225 g of desiccated thyroid, epinephrine caused little or no hyperglycaemia (Burn & Marks, 1925; Abbot & Van Buskirk, 1931). The amount of liver glycogen also affects the hyperglycaemic response to epinephrine; prolonged thyrotoxicosis depletes rabbit liver glycogen and then no hyperglycaemia follows epinephrine administration. In hypothyrodism, epinephrine produced a response smaller than normal.

Thyroid hormones affect hexose oxidation and hexose phosphorylation, directly and by modifications of the actions of other hormones. Oxidation of hexoses was accelerated by administered thyroid hormone, either through equal increases in both the phosphogluconate and the glycolytic pathways, or mainly through increased glycolysis; the route chosen may depend upon the degree of hyperthyroidism, low doses of hormone accelerating glycolysis mainly (Glock, McLean & White head, 1956) and depressing the phosphogluconate path (Dow & Allen, 1961). Hypothyroidism depressed glucose exidation via both paths (Dow & Allen, 1961). The mechanisms of the thyroid hormonal effects on glycolysis may be via one or more routes. An effect of thyroxine on the cytoplasmic acylphosphatase of rat liver and muscle has been demonstrated: administered hormone increases acylphosphatase activity, thyroidectomy decreases it, and then low doses of T_4 restore it (Harary, 1958). This enzyme hydrolyses 1,4-diphosphoglycerate to P_i and 3-phosphoglycerate; it acts as a rate-limiting ATPase, uncoupling glycolysis from phosphorylation, and

accelerates both glycolysis (by supplying P_i) and mitochondrial oxidation (by supplying ADP). And the activity of two enzymes of the glycolytic pathway, enolase and lactic dehydrogenase, were increased in the livers of thyrotoxic rats (Bargoni *et al.*, 1961), but they are probably not rate-controlling steps; the activities of a number of the glycolytic enzymes were decreased in hypothyroidism (Bargoni *et al.*, 1964). Lastly, the hormonal control of the generation of NADH and NADPH by mitochondria might also affect both glycolysis and the phosphogluconate pathway (Dow & Allen, 1961). Increased glucose uptake or oxidation, or both, have been observed in the muscles and in the livers of hyperthyroid animals, and also in cultures of animal cells, sperm, *Saccharomyces cerevisiae*, or *Acetobacter aerogenes* treated with thyroid hormones.

The rates of glucose utilization may also be affected through a hormonal control of hexose phosphorylation. Thyrotoxicosis raised the activity of intestinal phosphokinases (Nishikawara & Gabrielson, 1961), and the observed delays in the tolerance curves for glucose and galactose in this condition have been ascribed to rapid phosphorylation and intestinal absorption (Althausen, 1940). However, the importance of the phosphokinases in absorption has been questioned (Nishikawara & Gabrielson, 1961). The utilization rate of glucose, measured under constant intravenous load, is reported to be normal in thyrotoxicosis (Macho, 1958) and normal in hypothyroidism (Macho, 1961); in hypothyroidism, administration of thyroxine or DNP rapidly (4 hr) accelerated the utilization rate, suggesting that these two agents act similarly.

Intravenous glucose-tolerance tests gave high disappearance rates in hyperthyroid patients and low rates in hypothyroids; tolbutamide decreased blood glucose faster in hyperthyroids and slower in hypothyroids; and glucagon induced a lower glucose response in hyperthyroids (Lamberg, 1965). Orally administered D-xylose was normally absorbed by patients with thyrotoxicosis or myxoedema, but oral or intravenous D-xylose was excreted in the urine more rapidly in thyrotoxicosis, and less rapidly in myxoedema (Broitman *et al.*, 1964). Thyroid hormones potentiate insulin action; insulin-induced hypoglycaemia was increased in human thyrotoxicosis (Elrick, Hlad & Arai, 1961), and the uptake of glucose by rat adipose tissue was more sensitive to insulin after thyroxine was injected (Hagen, 1960).

Muscle contraction and creatine metabolism

Thyroid hormones control muscle contraction and creatine metabolism. In human hypothyroidism, skeletal muscles are larger and firmer than normal and contract slowly because of an abnormality in the contraction mechanism (Millikan & Haines, 1957). The clinical sign of the 'hung-up' reflex, with its slow relaxation, reflects this defect. In hyperthyroidism, muscle contracts at the normal rate, but performs work inefficiently (Plummer & Boothby, 1923). Clinically, 'thyrotoxic myopathy' (Thorn & Eder, 1946; Hed, Kirstein & Lundmark, 1958) reflects this defect. Thyroxine has a biphasic effect on muscular work in adrenalectomized rats, a low dose improving the work done per contraction, and a four-times-higher dose decreasing it (Ganju & Lockett, 1958).

The relation between muscle contraction and thyroid state thus seems clearer than in the case of some of the synthetic processes, probably because muscle contraction and relaxation depend more directly upon ATP supplied by mitochondrial oxidative phosphorylation. The other source of muscle ATP, the $\sim$P of phosphocreatine, can support only a few contractions and must itself be replenished from mitochondrial energy transformations. The skeletal muscles of thyrotoxic rats showed uncoupling (Johnson *et al.*, 1958), and those of thyrotoxic humans showed loose coupling (Ernster, Ikkos & Luft, 1959). In our terms, these mitochondria showed the action of the excessive amounts of thyroid hormones present. Other studies on apparently similar patients have shown normal muscle mitochondrial respiratory control, and either high controlled (State 4) and maximal (State 3) respiration (Stocker, Samaha & De Groot, 1966) or normal levels of respiration (Dow, 1967; Peter & Lee, 1967); because bovine serum albumin was used in the preparation and assay of the mitochondria, these results may show only the underlying enzymatic composition of the muscle mitochondria as contrasted with the action of the excessive amounts of hormone present *in situ*.

Heart muscle mitochondria are particularly susceptible to thyroid hormones (Bing, 1961). Clinically, this seems to be reflected in the high incidence of myocardial failure in thyrotoxicosis (the increased work load and decreased efficiency of contraction are an unfortunate combination), and in the lack of response of this form of failure to digitalis (which is more effective against mechanically induced defective contractile mechanisms).

Alterations in creatine metabolism usually involve muscle. A rationale for the observations is shown in Fig. 10. Clinically and experimentally, hyperthyroidism is accompanied by increased creatine excretion, and hypothyroidism by de-

creased creatine excretion. In hyperthyroidism, creatine synthesis was normal (Wilkins & Fleischmann, 1946). Injection of thyroxine rapidly depleted muscle phosphocreatine content, well before creatine excretion rose (Wang, 1946), suggesting a hormonal effect of phosphorylation. Thyrotoxic humans excreted creatine administered to them, or endogenously synthesized, in contrast to normal subjects, because of an inability to 'fix' creatine in their muscles, i.e. to resynthesize phosphocreatine from creatine and ATP (Shorr, Richardson & Wolff, 1933; Thorn, 1936). The defective creatine load-test is so characteristic that it has been used diagnostically.

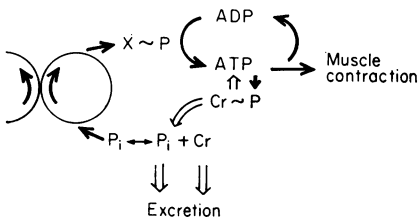

FIG. 10. Creatine and inorganic phosphate ion metabolism in relation to mitochondrial energy transfer. Creating phosphate (Cr ~ P), usually in equilibrium with the generated ATP, dissociates in hyperthyroidism to maintain the ATP level. The extra free creatine (Cr) and phosphate (P_i) are excreted. The P_i pool is augmented through inefficiency of oxidative re-esterification. Similarly, administering extra Cr in a load test augments the Cr-pool and subsequent excretion.

Vitamin metabolism

The thyroid hormones control the utilization of the water-soluble vitamins and their synthesis into coenzymes. The synthetic steps affected are usually energy-requiring condensations and phosphorylations. Most of the information is available on thyrotoxic subjects, where conditioned vitamin deficiences exist (Drill, 1943; Rawson, Rall & Sonenberg, 1955). Normal intake of vitamins is accompanied by deficiency symptoms because of increased demands or defective utilization.

Thiamine requirements are increased in hyperthyroid patients. The blood and liver vitamin contents were subnormal, and the excretion was higher than normal (Williams *et al.*, 1943). Tissue cocarboxylase content was low in hyperthyroid rats; it rose after thiamine was injected, but then fell more rapidly than in euthyroid animals, suggesting rapid destruction (Peters & Rossiter, 1939). *Pyridoxine* availability is limited in the tissues of hyperthyroid animals (Wohl *et al.*, 1960). Pyridoxal-5-phosphate content was low because of defective phosphorylation; conversely, it was high after thyroidectomy (Labouesse, Chatagner & Jollés-Bergeret, 1960). *Vitamin* B_{12} content was low in the tissues of thyrotoxic rats and hypothyroid female rats (Gershoff *et al.*, 1958; Kasbekar *et al.*, 1959); hormone administration raised renal B_{12} to normal in hypothyroid rats, and above normal in euthyroid rats (Okuda & Chow, 1961). *Ascorbic acid* content was low in the blood and tissues of thyrotoxic subjects. *Pantothenic acid* and CoA metabolism have been discussed under 'Lipid metabolism', and follow the same general pattern as the other water-soluble vitamins.

The thyroid hormones also control the synthesis of a fat-soluble vitamin. *Vitamin A* synthesis requires thyroid hormone. Both hypo- and hyperthyroid patients had poor dark adaptation (Wohl & Feldman, 1939). In hypothyroidism serum vitamin A was decreased because carotene was not converted to the vitamin; hormone treatment restored synthesis (Drill & Truant, 1947; Johnson & Baumann, 1947). In euthyroid animals, the hormone increased vitamin A synthesis, but prolonged treatment produced a severe resistant vitamin A deficiency (Portugal'skaya, 1961), another example of the hormone's biphasic effect.

Metabolism of inorganic ions

Phosphorus metabolism is strongly influenced by the thyroid state. Hyperthyroid patients are in negative phosphorus balance (Rawson *et al.*, 1955). Hypothyroid patients excreted large amounts of phosphate soon after administration of the hormone, probably because of increased phosphocreatine hydrolysis (Beaumont, Dodds & Robertson, 1940; Flach *et al.*, 1959). Phosphate contents were high, and ATP contents were low, in the soft tissues of thyrotoxic animals (Berg, 1937; Chatagner & Gautheron, 1960; Maley, 1957). The esterification of phosphate was slow in such tissues (Johnson *et al.*, 1958). In the bones of hyperthyroid patients, phosphate was turned over abnormally rapidly (Hernberg, 1960) probably in conjunction with the changes in calcium.

Calcium metabolism also changes in thyroid disease. Calcium turnover in bones is accelerated in hyperthyroid patients, and becomes normal with the thyroid state upon treatment (Krane *et al.*, 1956). Calcium accumulation was more striking in the livers than in the bones of thyroxine-treated rats; the capacity of liver mitochondria to store Ca^{++} may be involved. In hypothyroid rats, calcium incorporation into bone was decreased (Lengemann, Wasserman &

Comar, 1960) but in hypothyroid humans, hormone administration did not raise calcium excretion rapidly, although it raised phosphate excretion (Beaumont *et al.*, 1940), perhaps because the hormone acts more directly on phosphate metabolism.

Magnesium metabolism depends upon the thyroid state, and vice versa. Mg^{++} and thyroid hormones are antagonists *in vivo* and *in vitro* when mitochondrial function is measured (see Hoch, 1962b). Myxoedematous patients excreted large amounts of Mg^{++} in their urine promptly after hormone administration (Tapley, 1955). The plasma magnesium content was low in hyperthyroidism and high in hypothyroidism; Mg balance was positive in hyper- and negative in hypothyroidism; total and cellular exchangeable Mg^{++} was strikingly low in hypothyroidism but normal in hyperthyroidism (Jones *et al.*, 1966). The effect of administering magnesium salts upon thyrotoxicosis is controversial, some finding a decrease in BMR and heart rate (Hueber, 1939), others finding no decrease in BMR nor change in negative nitrogen and phosphate balances Wiswell, 1961).

Decreased exchangeable *potassium* (Munro, Renschler & Wilson, 1958; Wayne, 1960) and hyperkalaemia and hyperkaluria (Boekelman, 1948) have been reported in thyrotoxicosis. Occasionally periodic muscular paralysis is associated with hyperthyroidism.

Temperature regulation

The thyroid hormones are involved in the control of body temperature. About 60% of the energy liberated by mitochondrial oxidations is normally converted to a chemically utilizable form, the other 40% being liberated as heat and thereby maintaining the body temperature of homeotherms. In hyperthyroidism heat production is raised by two factors: the increased rate of oxidation and the decreased efficiency of energy conversion. Usually the excess amounts of heat can be dispelled by physiologic compensations such as flushing, sweating and increased circulation; many of the clinical characteristics of hyperthyroid patients arise from these compensations.

Thyrotoxic crisis or storm can be viewed as a failure of compensation due to increased heat production through a further loss of mitochondrial efficiency. Body temperatures rise sharply to 107°F or more, muscle tone is lost, liver damage (? mitochondrial) is severe and the patients may die. Body refrigeration may remove enough heat to save the situation, but therapeutic measures to provide adrenocortical hormones and to antagonize adrenomedullary hormones have also been used with success.

The experimental induction of the acute hyperthermia that is seen clinically in thyroid crisis provides an insight into how the thyroid hormones act physiologically. Administering large doses of thyroid hormone to animals usually produces a loss of weight and an apathetic death, not a hyperthermic crisis. Much smaller doses of hormone, however, can produce fatal hyperthermia in conjunction with the administration of an agent that acts on mitochondrial oxidative metabolism. Among such compounds are the uncoupling agents, like dinitrophenol (Hoch, 1965a) dinitro-*o*-cresol (Barker, 1946) and methylene blue (Alwall, 1936); phosphate ions given by infusion (Roberts *et al.*, 1956); and antipyretic agents, like sodium salicylate (Hoch, 1965a). A dose of salicylate too small to raise the BMR in a normal rat raises the BMR sharply in a midly hyperthyroid rat; one-quarter of the normally lethal dose of salicylate rapidly induces a fatal hyperthermia in such hyperthyroid rats. With dinitrophenol, this phenomenon can be shown to arise from an exaggerated sensitivity of mitochondria to the uncoupling agent, induced by thyroxine treatment (Hoch, 1968c). Whether the clinical phenomenon has a similar basis remains to be seen. The association of thyroid crisis in thyrotoxic patients with infections that cause fever (Means, De Groot & Stanbury, 1963) may be another example of an *in vivo* synergism.

In hypothyroidism, heat production is diminished by the depressed rate of mitochondrial oxidations. Again there are physiologic compensations to preserve body heat, and the skin is cold, circulation is slow, and cold is poorly tolerated. Body temperatures may be below normal. Infections that normally elicit fever may not raise the hypothyroid patient's temperature at all, or at least not above normal. Occasionally a fatal hypothermia may supervene, the so-called *myxoedema coma*, in which body temperature can no longer be maintained, and has been reported as low as 74°F. Experimentally the calorigenic response of hypothyroid rats to an administered uncoupling agent is subnormal (Hoch, 1965b), because their mitochondria are subnormally sensitive (Hoch, 1967, 1968c). Administered thyroxine rapidly raises mitochondrial responses, and the efficiency of such treatment clinically may be evidence for a common basis for the hypothermia.

Effects of hormones and drugs

The effects of a number of hormones and

drugs depend upon the thyroid state of the subject, as has been mentioned above in connection with specific systems. In general, hyperthyroidism exaggerates and hypothyroidism minimizes the changes seen after administration of the agent. Physiologically, the clearest example is that of the *catecholamines*. The relationship is so striking that some have concluded that the apparent peripheral effects of thyroxine are actually effects of epinephrine (Brewster *et al.*, 1956), but there is evidence against so sweeping a claim (see Hoch, 1962b). The biochemical basis of the observed interdependences may be the inactivating effects (or actions?) of thyroid hormones upon the enzymes that normally inactivate the catecholamines themselves. The catechol-*o*-methyl-transferase (D'Iorio & Leduc, 1960), the amine oxidases (Zile & Lardy, 1959), and a peroxidase system (Klebanoff, 1959) have been studied, but it is still difficult to assign physiologic relevance to the mechanisms. Another enzymatic system under scrutiny in this regard involves the formation of cyclic-3′,5′-AMP, the mediator of many of the catecholamine effects. Thyroid hormones have been suggested to control the activation of the lipase in adipose tissue via a mechanism involving cyclic-3′,5′-AMP (Fisher & Ball, 1967). Yet other possible routes are the control the thyroid hormones exert over ATP availability, since ATP is the only source of cyclic-3′,5′-AMP; and the sensitivity of mitochondria. Adrenochrome and thyroxine act synergistically on mitochondria *in vitro* (Park, Meriwether & Park, 1956). The glycogenolytic and hyperglycaemic, the lipolytic, the inotropic, and the calorigenic effects of epinephrine all depend upon the thyroid state (see Ellis, 1956; Brodie *et al.*, 1966; Goodman & Bray, 1966). The abnormally slow pulse rate after epinephrine administration in hypothyroid patients, and the rapid rate in hyperthyroid subjects, have been used diagnostically, although caution is advised in hyperthyroids (Goetsch & Ritzmann, 1934).

The dependence of the calorigenic effect of epinephrine upon the thyroid state is an example of the generality that the thyroid state controls the response of the body to calorigenic substances. Excessive rise in BMR is seen in hyperthyroidism, and little or no rise is seen in hypothyroidism, after the administration of glucagon (Davidson, Salter & Best, 1960), nitrophenols, salicylates (Hoch, 1965a, b), chloropromazine (unpublished data), and 'febrile toxins' (above). The only exception seems to be the enhanced sensitivity of hypothyroid subjects to the thyroid hormone itself.

The adrenal cortical hormones and insulin may have synergistic effects with the thyroid hormones on a physiological level, but these effects involve different rates of catabolism and production of all three groups of hormones, as well as interactions in the tissues.

Other features

There are several clinical features of hyperthyroidism and hypothyroidism that are not readily reduced to manifestations of changed cellular energy transfer. These features fall into two categories. First, there are those that arise from mechanisms not due directly to the changed amounts of thyroid hormones in the tissues, but to phenomena associated with the primary defect in thyroid hormone production. Thus, exophthalmos is one of the classical signs in the Merseburg triad in hyperthyroidism but it is not produced by hormone administration (Means *et al.*, 1963). A pituitary factor, possibly associated with thyrotropin, may be responsible (Loeb & Friedman, 1932). The frequent persistence of exophthalmos after the therapeutic restoration of euthyroidism speaks for such a secondary relationship.

In the second category are those clinical features that may (and indeed seem to) arise from changed amounts of tissue thyroid hormone, but that are not reducible to cellular phenomena because we don't know enough yet. The hyperirritability of the nervous system in hyperthyroidism, and its opposite in hypothyroidism, may be presenting symptoms clinically. The involvement of ATP in nerve conduction and in resynthesis of acetylcholine at the myoneural junction, and the involvement of K^+, Ca^{++} and Mg^{++} in neural events make it likely that hormone-induced defects in energy transfer and ion accumulation will affect the nervous system, but just how changes in nerve function relate to 'nervousness' presents complex problems not yet conclusively approached. Similarly, the presenting abnormality of 'myxoedema' seems to be a defect in mucopolysaccharide metabolism that leads to excessive deposition, somehow and presumably dependent upon insufficient thyroid hormone.

The thyroid hormones obviously control growth, development, and the striking structural and chemical changes in Anuran metamorphosis. In general terms we may say these processes must depend upon available 'energy' but our lack of knowledge of the details of the energy-dependent steps precludes a mechanistic interpretation at present. In this area, however, the recent evidence that the thyroid hormones con-

trol mitochondrial energy metabolism directly and promptly, and thereby regulate protein synthesis, offers promise of new and important information on biologic and medical problems.

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